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中国西南地区地图衣属一新种——四川地图衣

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摘要：本研究以中国西南地区为研究区域，采用表型特征（形态学、解剖学和化学特征）和基因型特征（ITS 序列）相结合的方法，对地图衣属（*Rhizocarpon* Ramond ex DC.）地衣物种进行分类学研究，明确了地图衣属部分物种的分类地位，初步探讨了该属物种表型与基因型的对应关系。通过本研究，我们发现了该属 1 新种——四川地图衣（*Rhizocarpon sichuanense* Y. M. Zhang, L. Hu & W. C. Wang, sp. nov.），该新种与相近种黑灰地图衣（*Rhizocarpon cinereonigrum* Vain.）和中华地图衣（*Rhizocarpon sinense* Zahlbr.）的主要区别是：地衣体裂片较为分散，表面呈龟裂或者亚鳞叶状；下地衣体黑色，明显；孢子较大，（27－）32－42.5×12.5－17.5（–20）μm，TLC 检测含有巴巴酸。在系统发育中，该新种与黑红地图衣[*Rhizocarpon badioatrum* (Flörke ex Spreng.) Th. Fr.]在同一个进化分枝上，但是黑红地图衣的地衣体裂片连续不分散，孢子明显较小（23－36×13－16 μm），地衣体中含有地弗地衣酸或不含化学物质。通过系统发育分析，结果表明：（1）该新种——四川地图衣属于褐地衣体亚属中的 Badioatrum group；（2）仅依据解剖学特征（孢子的大小和分隔类型）对地图衣属部分类群的划分存在不合理性，还需结合化学特征进行综合分析。该文提供了新种的分类学描述以及形态、解剖和化学的高分辨率图片。利用新种的 ITS 序列构建的系统发育树，为建立更趋自然合理的分类系统提供基础数据。此外，本文对世界范围内褐地衣体亚属中子囊孢子为棕色 1 隔的类群（Badioatrum group）编写了详细的物种检索表，为该类群的进一步研究提供了资料支持。

关键词：巴巴酸，地图衣科，地衣型子囊菌，地衣型真菌，分类学

中图分类号：Q948.2；Q949.34 **文献标识码：**A **文章编号：**

Rhizocarpon sichuanense, a new species from Southwest China

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Abstract: In this paper, we study in *Rhizocarpon* Ramond ex DC. species by the method of combining phenotypic characteristics (morphology, anatomy and chemistry) with genotypic characteristics (ITS sequence). The aims of this present paper are to clarify the status of some species and explore the relationship between phenotype and genotype in *Rhizocarpon*. During the study on *Rhizocarpon* species from southwest China, *R. sichuanense* Y. M. Zhang, L. Hu & W. C. Wang sp. nov.

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is described as a new species in this genus. The new species is morphologically similar to *R. cinereonigrum* Vain and *R. sinense* Zahlbr., which also produces brown thallus, medulla I-, 8-spored ascus and brown ascospores with 1-septa. But it can be distinguished by the scattered, areolate to subsquamulose thallus, a distinct black prothallus, large ascospores [(27–)32–42.5 × 12.5–17.5(–20) μm] and the presence of barbatic acid in the thallus. In phylogenetic trees, it is also similar to *R. badioatrum* (Flörke ex Spreng.) Th. Fr. which can be distinguished by the more continuous thallus with diffractaic acid or without secondary products, and the smaller spores (23–36 × 13–16 μm) than this new species. The phylogenic results are as follows: (1) The new species *R. sichuanense* belongs to Badioatrum group which belongs to *R.* subg. *Phaeothallus* ; (2) It is unsuitable to classify some groups only based on anatomical characteristics (spore size and separation type) in *Rhizocarpon*, the chemical characteristics should also be considered. In this paper, the achievement of ITS sequences provide the basic data for an establishment of a more reasonable and natural classification system. In addition, we provide high resolution pictures of morphological, anatomical and chemical characteristics as well as detailed descriptions of the new species. The key to the species of *R.* subg. *Phaeothallus* with 1-septa brown spore (Badioatrum group) in the world is also provided, which will provide basic data for the further studys in this group.
Key words: barbatic acid, Rhizocarpaceae, lichenized ascomycetes, lichenized fungi, taxonomy

地图衣属由 Candolle 于 1805 年建立，该属的主要特征是：地衣体生长型为壳状，呈裂缝至鳞叶状，裂片连续或分散，表面黄绿色、白色、灰色至深棕色；子囊盘黑色，网衣型，盘缘较窄，不明显或明显加厚；果壳外缘通常着色较深，内部着色较浅；子实上层棕色至蓝绿色，K±紫红色；子囊内含 1-8 孢子，子囊顶部与碘反应后，子囊外壁呈深蓝色，内壁仅在顶部略有着色，称为地图衣型子囊顶器（*Rhizocarpon*-type）；子囊孢子无色至棕色或墨绿色，1 至多隔或呈亚砖壁至砖壁型，晕圈明显；囊层基深棕色（Hafellner, 1984）。1956 年，Runemark 在研究欧洲地图衣属黄绿色类群地衣时，将孢子 1 隔、长度为 9–18 μm 的物种归为 Superficiale group；孢子 1 隔、长度为 18–32 μm 的物种归为 Alpicola group；孢子砖壁型、髓层 I－的物种归为 Viridiatrum group；孢子砖壁型、髓层 I+的物种归为 Geographicum group（Runemark, 1956）。1967 年，Thomson 根据地衣体是否含有地图衣酸（rhizocarpic acid）将地图衣属划分为 2 个亚属：即地衣体为黄绿色，含有地图衣酸的物种归为地图衣亚属（*Rhizocarpon* subg. *Rhizocarpon*），地衣体为非黄绿色（白色、灰色至棕色），不含有地图衣酸的物种归为褐地衣体亚属（*R.* subg. *Phaeothallus*）（Thomson, 1967）。Poelt（1988）又将隶属于地图衣亚属的物种进一步做了总结，汇编成一个较为完整的检索表。随后，Fryday（2000，2002）和 Ihlen（2004）在研究欧洲地图衣属非黄绿色类群时，根据孢子的特征将褐地衣体亚属中孢子棕色、1 隔的物种归为 Badioatrum group；孢子无色、砖壁型的物种归为 Obscuratum group；孢子无色、1 隔的物种归为 Hochstetteri group。

随着分子生物学的发展，Mccune 等（2016）和 Davydov 等（2017）通过表型结合基因型（ITS）的方法对阿拉斯加半岛和西伯利亚阿尔泰山地区的地图衣属进行系统发育学研究时，发现了新种 2 个：*R. quinonum* McCune, Timdal & Bendiksby、*R. smaragdulum* Davydov & Yakovch.，新记录种 2 个：*R. atroflavescens* Lynge 和 *R. norvegicum* Räsänen。Roca-Valiente（2013 年）通过多基因联合分析的方法对地图衣亚属中的 Geographicum group 进行了系统发育学研究，结果表明 Geographicum group 中的地衣物种表型特征与基因型特征并不完全符合，仅依据表型特征的分

类学研究不足以反映该类群的自然演化特征及系统发育关系。因此亟需通过表型和基因型相结合的方法对地图衣属进行系统的分类学研究。

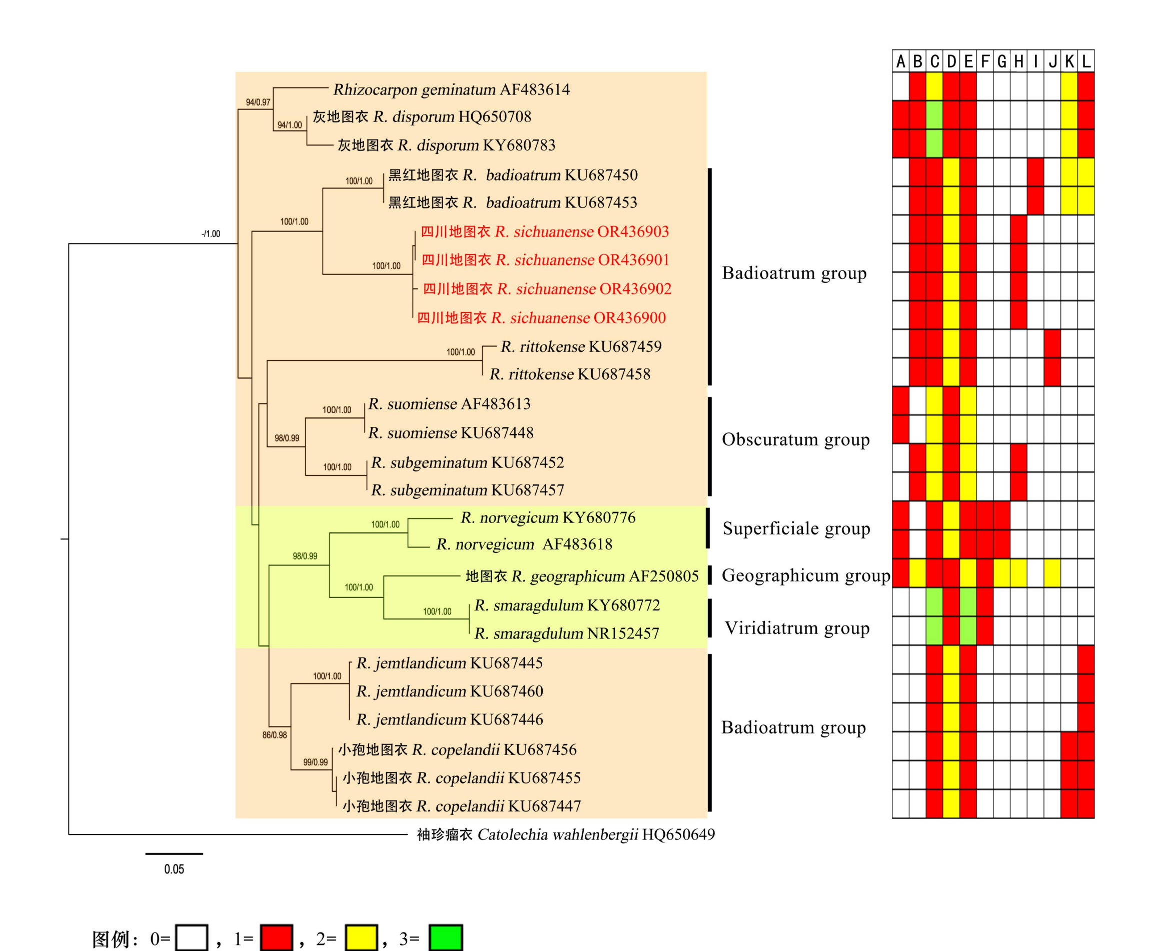
目前全球报道地图衣属约有 230 种，该属物种主要分布在高原、山地等气候较为寒冷的地区（Lücking et al., 2016；Davydov &Yakovchenko, 2017；Kalb & Aptroot, 2017；Paukov et al., 2017；Kondratyuk et al., 2018；Fryday, 2019；Elix & McCarthy, 2019；McCarthy et al., 2020；Spribille et al., 2020）。中国仅报道地图衣属地衣 47 种，主要集中在中国的西南（云南、四川、贵州、湖北、西藏）和西北地区（新疆、青海、甘肃、陕西、山西、内蒙古西部）（魏江春，1991；阿巴斯和吴继农，1998；Aptroot & Sparrius, 2003；Sérusiaux et al., 2003；Golubkov et al., 2009；李雪等，2013；Zhao et al., 2013；Mahire et al., 2015；Wang et al., 2016, 2015a, b, c；Gulina & Anwar, 2019；Hu et al., 2020；Bi et al., 2022）。我国地域辽阔，生物多样性高，蕴藏着十分丰富的地衣资源，但研究基础较为薄弱，据估计，我国已知地衣型真菌种数仅占估计种数的 8.5%，尚有 91.5％的物种有待调查研究（魏江春，2018）。长期以来我国学者对地图衣属的研究多为零星报道，且研究方法主要依据经典分类学（表型特征）方法，因此利用表型结合基因型的研究方法对中国地图衣属地衣开展分类学研究具有重要意义。

本研究采用表型（形态学、解剖学和化学）结合基因型（ITS 序列）的方法对地图衣属地衣物种进行分类学鉴定，目的是明确地图衣属部分物种的分类地位，为建立更趋自然合理的分类系统提供数据支持；探讨该属部分物种表型与基因型的对应关系，为进一步研究地图衣属提供可参考资料；世界范围内 Badioatrum group 物种检索表的整理为该类群的研究提供基础资料。

1 材料与方法

本研究所用的地衣标本采自四川省甘孜藏族自治州和凉山彝族自治州，保存在山东师范大学植物标本馆（SDNU）和中国科学院昆明植物研究所地衣标本馆（KUN-L）中。

利用体式显微镜（Olympus SZX16）对地衣的外形特征（包括地衣体的颜色、厚度，子囊盘的形态、颜色和大小等）进行研究并拍照记录，利用光学显微镜（Olympus BX61）对标本的解剖学特征进行观察并拍照记录，如子实上层、子实层、囊层基和果壳的颜色，厚度以及晶体分布情况。皮层和髓层的显色反应则是利用K试剂（10% 氢氧化钾溶液），C试剂（次氯酸钠饱和溶液），IKI试剂（10% Lugol’s碘液）和P试剂（对苯二胺在95%乙醇中的饱和溶液）进行检测。地衣的次生代谢产物则是利用薄层层析法（thin layer chromatography techniques, TLC）在B系统中进行检测（Culberson, 1972；Culberson & Kristinsson, 1970）。



A. 髓层碘反应：0=无反应，1=变蓝；B. 子实上层 K 反应：0=无反应，1=变紫红色，2=有或无紫红色；C. 子囊内孢子数目：1=8 孢，2=2 孢，3=1 孢；D. 孢子类型：1=砖壁型，2=具 1 横隔；E. 孢子颜色：1=无色，2=棕色/绿色，3=幼时无色成熟时颜色加深；F. rhizocarpic acid：0=无，1=有，2=有或无；G. psoromic acid：0=无，1=有，2=有或无；H. barbartic acid：0=无，1=有，2=有或无；I. diffractic acid：0=无，1=有，2=有或无；J. gyrophoric acid：0=无，1=有，2=有或无；K. norstictic acid：0=无，1=有，2=有或无；L. stictic acid：0=无，1=有，2=有或无。

每个分支的左侧数值表示自展值(BS)，右侧数值代表贝叶斯后验概率(PP)，本结果仅显示自展值大于75或后验率大于0.95的数值。该系统发育树中的外类群为地图衣科中的瘤衣属。黄色背景为地图衣亚属的物种，橙色背景为褐地衣体亚属的物种，红色标记的序列由本研究新产生。核苷酸替代率为0.05。

The number in each node represents bootstrap support (BS) and posterior probability (PP) values. BS values≥75 (left) and PP values ≥ 0.95 (right) were plotted on the branches. The phylogenetic tree was rooted to *Catolechia* of Rhizocarpaceae. The sequences in yellow background shows the species belong *Rhizocarpon* subg. *Rhizocarpon*, the orange background shows the species belong to *R.* subg. *Phaeothallus*, the sequences of taxa in red color were newly generated for this study. Scale in 0.05 substitution per site.

图 1 基于 ITS 序列构建的地图衣属系统发育树

Fig.1 Phylogenetic tree of *Rhizocarpon* based on ITS sequences

利用Sigma-Aldrich DNA提取试剂盒按照操作说明提取每份标本的基因组，提取的DNA模板可以长期保存于-20℃。使用真菌特异性引物ITS1F（Gardes & Bruns, 1993）和ITS4（White et al., 1990）对每份样本的ITS序列进行PCR扩增，PCR设置程序如下：首先，95℃ 2 min；然后，35个循环，包括：94℃ 20 s，53℃ 60 s，72℃ 2 min；最后72℃ 15 min，最终产物在4℃进行保存。本研究获得的PCR产物送青岛博尚生物科技有限公司纯化和测序。测序获得的单端序列使用SeqMan（Swindell & Plasterer, 1997）进行校正和拼接，在GenBank中下载地图衣属相关序列，构建系统发育树，用MAFFT v. 7（Katoh et al., 2009）进行序列比对，使用Gblocks V0.19b（Talavera & Castresana 2007）去除比对过程中产生的模糊序列。通过jModelTest 2（Darriba et al., 2012）检验选择贝叶斯法（Bayes, BI）最佳分析模型，最大似然法（maximum likelihood, ML）使用RAxML v. 8.2.6（Stamatakis, 2014），以1000次重复计算自展法（bootstrap）评估各分支的支持率，然后在Cipres Science Gateway（http://www.phylo.org）网站运行最大似然法和贝叶斯法分析。运算生成的进化树文件使用FigTree v.1.4.3查看和调整。自展值（BP）大于75%或贝叶斯后验概率（posterior probability, PP）大于0.95的分支认为可信。

2 结果与分析

2.1 分子分析结果

本研究新产生的4条地图衣属ITS序列与GenBank已下载的23条参考序列比对、去除模糊序列后形成包括27个物种的546个位点矩阵。比对ML树与BI树，有相同的拓扑结构，因此文中仅展示ML树，将BI树的后验概率标记在节点的分支处的自展值之后（图1）。

本研究中的新物种已在Fungal Names网站（https://nmdc.cn/fungalnames）注册，新产生的序列已上传保存至GenBank（https://www.ncbi.nlm.nih.gov/genbank）。

2.2 物种描述

四川地图衣（图 2）

***Rhizocarpon sichuanense* Y. M. Zhang, L. Hu & W. C. Wang sp. nov.** **图 2 (A-J)**

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Diagnosis: This species is similar to *Rhizocarpon cinereonigrum* Vain. but differs by the large brown ascospores, areolate to subsquamulose thallus, grey-brown areolae, and the presence of barbatic acid.

Etymology: The epithet ‘sichuanense’ refers to the Sichuan province, where this species was found.

Typus: China, Sichuan, Garze Tibetan Autonomous Prefecture, Kangding City, Ertaizizhongqiao Co., 30°2'34.24"N, 101°49'53.27"E, 3752 m alt., on rock, 29 April 2021, Ling Hu, et al., 20210551 (SDNU–holotypus).

Description: Thallus crustose, areolate to subsquamulose, generally up to 5 cm diam, sometimes larger, areoles brown to grey-brown, flat to slightly convex, matt, scattered on prothallus, round or angular, up to 0.75 mm diam, 0.1–0.5 mm thick, cracks usually present on the areolae, epruinose to faintly pruinose. Upper cortex brown, dull, smooth, without an epinecral layer, 12–20 μm high, containing crystals. Lower cortex black brown, without crystals. Photobiont layer continuous, 65–95 μm high; photobiont chlorococcoid, algal cells 10–15 μm diam. Prothallus distinct, black.Prothallus well developed, black, distinct between the areolae and along the margin;

Apothecia black, lecideine, 0.2–0.9 mm diam, round or angular between the areoles, sessile on the black prothallus; disc flat to concave, scabrid, epruinose; proper margin 50–100 µm thick, persistent, above or the same level as the disc, concolourous with the disc, epruinose to faintly pruinose, sparingly shiny; exciple 50–100 µm thick, deep brown to black at the rim; inner part red-brown, containing crystals dissolved in K, the radiating hyphae 4–6 µm wide; hymenium colorless, 80–135 µm tall, paraphyses septate, branched and anastomosing c. 2.5 µm thick in mid-hymenium and 3–4(–5) µm thick apically; epihymenium pale brown to deep brown, K+ pale violet-red, to 25 µm thick, without crystals, K–; hypothecium black brown, without crystals, K–; asci clavate, *Rhizocarpon*-type, 8-spored; ascospores soon becoming dark brown, 1-septate, ellipsoid or oblong, halonate, (27–)32–42.5 × 12.5–17.5(–20) µm, length/width ratio (1.4–)1.7–2.1(–2.2).

Chemistry: Medulla I–, K–, C–, KC–, P–; barbatic acid (TLC).

Distribution and habitat. The new species is known from Sichuan Provinces, on huge granite stones, between 2731 m and 3752 m altitude, growing with *Rhizocarpon geographicum*, *Dermatocarpon* sp. and *Aspicilia* sp. It is only known in China up to now.

Additional specimens examined. China, Sichuan, Garze Tibetan Autonomous Prefecture, Luding County, Mt.Yajiageng, on the rock, 29°54'10.19"N 101°59'59.64"E, alt.3946 m, 27 April 2021, Ling Hu et al., 20210102 (SDNU); China, Sichuan, Liangshan Yi Autonomous Prefecture, Huili County, on the rock, 26°47'22.89"N 102°12'18.58"E, alt.3648 m, 23 April 2021,Ling Hu et al., 20210837 (SDNU). China, Sichuan, Liangshan Yi Autonomous Prefecture, Puge County, on the rock, 27°35′5.63″N, 102°22′47.40″E, alt.3622 m, 13 September 2021, X.Y. Wang et al., XY21-418 (KUN 80845)

Remarks. *Rhizocarpon sichuanenseis* characterized by its saxicolous, scattered grey-brown and areolate to subsquamulose thallus, a distinct black prothallus, large 1-septate brown ascospores and the presence of barbatic acid. *R. cinereonigrum* is similar to this new species but the fomer has smaller ascospores (25–36 µm long) and produces stictic acid in thallus (Thomson, 1977). Phylogenetic trees (Fig. 2) indicate that the new species is close to *Rhizocarpon badioatrum* (Spreng.) Th. Fr., also has brown epihymenium K+ violet and 1-septate brown ascospores, but *R badioatrum* has a dark brown and more continuous thallus with diffractaic acid or without secondary products (Timdal & Holtan-Hartwig, 1988). Among all of the non-yellow *Rhizocarpon* species with 1-septate brown ascospores, *R. sichuanense* has the largest ascospores (32–42.5 µm long), *R. badioatrum* and *R. cinereonigrum* have smaller ascospores (25–36 µm long); and other species are even smaller (less than 30 µm long) (Fletcher et al., 2009). Therefore, combined with the above-mentioned morphological characteristics, we identified *R. sichuanense* as a new species.

主要特征：该种与*Rhizocarpon cinereonigrum* Vain.相似，但是区别在于该种的孢子较大，地衣体龟裂至亚鳞叶状，裂片表面灰棕色，次生代谢产物为巴巴酸。

词源：新种的种加词‘sichuanense’指该种模式发现的地点为四川省

主模式：中国，四川省，甘孜藏族自治州，康定市，二台子中桥，30°2'34.24"N，101°49'53.27"E，3752 m，石生，2021.04.29，胡玲等，20210551（SDNU－holotypus）。

形态学描述：地衣体壳状，表面龟裂至鳞叶状，直径通常可达 5 cm。地衣体裂片棕色或灰棕色，平整或者略有凸起，在前地衣体上分散排列，呈圆形或具棱角，直径可达 0.75 mm，厚度为 0.1–0.5 mm，裂片表面粗糙，通常有细微裂缝分布，无粉霜或略有轻微粉霜分布。上皮层棕色，暗淡，平整，无胶质层分布，高度为 12–20 µm，有晶体分布。下皮层深棕色，无晶体分

布。藻层连续，高度为 65–95 µm；共生藻为球形绿藻，藻细胞直径为 10–15 µm。前地衣体黑色，发育良好，分布在地衣体裂片间或裂片边缘。

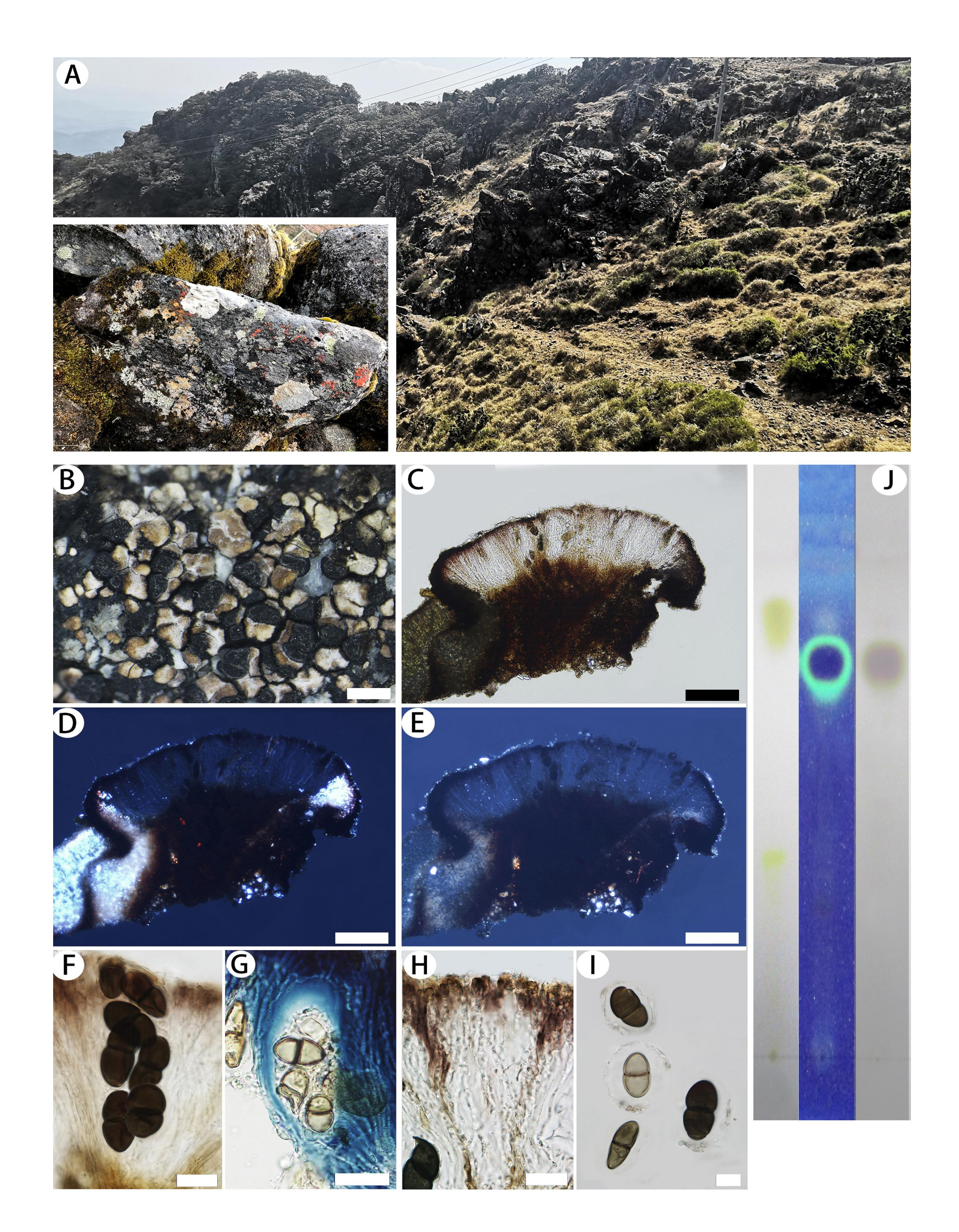
子囊盘黑色，网衣型，直径为 0.2–0.9 mm，圆形或具棱角，分布在地衣体裂片之间，贴生于黑色的前地衣体之上；盘面平整至凹陷，略粗糙，无粉霜；果壳较厚，明显可见，厚度为 50–100 µm，比盘面略高或与盘面同高度，颜色与盘面相同，均为黑色，无或略有粉霜分布，极少有光泽；果壳外缘着色较深，为深棕色至黑色，果壳内部着色较浅，为红棕色，有可溶于 K 试剂的晶体分布，果壳内部菌丝呈辐射状，宽度为 4–6 µm；子实层通常无色，高度为 80–135 µm；侧丝分隔，粘合且分枝，侧丝中部菌丝细胞约为 2.5 µm 厚，侧丝顶部菌丝细胞 3–4（–5）µm 厚；子实上层浅棕色至深棕色，K 反应呈浅紫罗兰色，厚度可达 25 µm，无晶体分布，K–；囊层基棕色，无晶体分布，K–；子囊棒状，子囊顶器类型为地图衣型（*Rhizocarpon*-type），子囊内含 8 个孢子；子囊孢子成熟时快速变成深棕色，具有 1 横隔，横隔处略有缢缩，椭圆形至长椭圆形，具晕圈，（27–）32–42.5×12.5–17.5（–20）µm，长/宽为（1.4–）1.7–2.1（–2.2）。

化学特征：髓层I–, K–, C–, KC–, P–；TLC法检测到巴巴酸（barbatic acid）。

分布：该种采自四川省，通常分布在巨大的花岗岩上，海拔范围在2731 m至3752 m之间，与*Rhizocarpon geographicum*、*Dermatocarpon* sp. 和*Aspicilia* sp. 等地衣物种生长在一起。目前为止，该种仅见于中国。

其他引证标本：中国，四川省，甘孜藏族自治州，泸定县，雅家梗山，3946 m，石生，2021.4.27，胡玲等，20210102（SDNU）；中国，四川省，凉山彝族自治州，会理县，3648 m，石生，2021.4.23，胡玲等，20210837（SDNU）；中国，四川省，凉山彝族自治州，普格县，3622 m，石生，2021.9.13，王欣宇等，XY21-418（KUN 80845）。

讨论：该新种主要特征是地衣体石生，龟裂至亚鳞叶状，裂片分散分布，表面灰棕色；下地衣体黑色，明显；孢子较大，具1横隔，地衣体中的次代产物为巴巴酸。*Rhizocarpon cinereonigrum*与该新种相似，但是前者的孢子通常较小（长度为25–36 µm），且地衣体中的次代产物为斑点酸（stictic acid）（Thomson, 1977）。系统发育分析结果表明该新种与*R. badioatrum* (Spreng.) Th. Fr.进化距离较近，后者也具有棕色的子实上层，且K反应也为紫红色，孢子也为棕色具1横隔，但是*R. badioatrum*的地衣体裂片通常连续分布，且地衣体颜色较深，为深棕色，前地衣体通常分布在地衣体边缘，孢子较小，23–36×13–16 µm，次代产物为地弗地衣酸（diffractaic acid）（Timdal & Holtan-Hartwig, 1988）。此外，*R. sinense* Zahlbr.的孢子也较大（30–40×10–15 µm），且具 1 横隔，但是该种地衣体着色较深，为深棕色，表面具光泽，有胶质层分布，次代产物为三苔色酸（gyrophoric acid）（C+红色），子囊盘为埋生，孢子为橄榄棕色（Zahlbruckner, 1930）。



A. 模式种产地，康定市二台子中桥，分布在 318 国道两侧山丘中的岩石上；**B**. 地衣体灰棕色，裂片分散，呈亚鳞叶状；**C**.子囊盘纵切片；**D**. 果壳里有晶体分布；**E**. 果壳内晶体溶于 K 试剂；**F**. 子囊；**G**. 子囊顶器碘反应；**H**. 子囊孢子棕色 1 隔，具晕圈；**I**. 侧丝分枝且粘合；**J**. TLC 结果示巴巴酸（左为标样在日光下硫酸显色；中为新种在 365 nm 光下硫酸显色；右为新种在日光下硫酸显色）。标尺：**B** = 0.5 mm；**C–E** = 100 µm；**F–I** = 20 µm。

A. Type locality, Kangding City, Ertaizizhongqiao County, typical habitat is distributed on stones in the hills off Highway 318; **B**.Grey-brown thallus with scattered areoles, apothecia and subsquamulose areoles; **C**. Vertical sections of apothecia in water; **D**. Showing crystals in excipulum refracting polarized light; **E.** Showing crystals dissolved in K; **F**. Ascus; **G**. Amyloid reaction of ascus; **H.** 1-septate brown ascospores with haloe; **I.** Showing branched and anastomosing paraphyses; **J.** TLC results show barbatic aicd. (On the left, the standard sample is sulfuric acid color under sunlight; in the middle, the new species is sulfuric acid color under 365 nm light; on the right, the new species is sulfuric acid color under sunlight). Scales: **B** = 0.5 mm; **C–E** = 100 µm; **F–I** = 20 µm.

图 2　四川地图衣（SDNU 20210551）

Fig.2　Holotype of *Rhizocarpon sichuanense* sp. nov. (SDNU 20210551)

目前，在褐地衣体亚属里，孢子棕色且具1横隔的物种（Badioatrum group）中，孢子最大的是*R. sichuanense*（长度为32–42.5 µm），其次为*R. sinense*（长度为30–40 µm），*R. badioatrum*和*R. cinereonigrum*的孢子相对较小（长度为25–36 µm），而其他物种的孢子更小（长度小于30µm）（Fletcher et al., 2009）。因此，根据以上的形态学、解剖学、化学以及分子生物学结果，我们认为该新种成立。

此外，本研究通过结合表型与基因型特征进行分析发现，在亚属水平的划分上，地图衣亚属和褐地衣体亚属的物种并不能形成单独的进化分枝，而是前者包含于后者之中，由此可见Thomson（1997）仅依据地衣体的颜色以及是否产生rhizocarpic acid对地图衣属进行划分并不合理，同样的研究结果在Mccune等（2016）和Davydov等（2017）中均有发现。在种间或者类群（group）水平的划分上，我们发现地图衣亚属的物种表型与基因型特征基本符合，即根据孢子的大小、分隔类型以及髓层碘反应划分的Superficiale group、Viridiatrum group和Geographicum group这个3个类群的物种各自形成了单独的进化分枝。然而，在褐地衣体亚属中，尤其是Badioatrum group的物种并没有聚集到一起，而是*R. sichuanense*与*R. badioatrum*，*R. copelandii*与*R. jemtlandicum*各自聚成一个进化分枝，*R. rittokense*单独形成一个分枝，由此可见，对褐地衣体亚属尤其是Badioatrum group的划分，不能仅依据孢子的颜色和分隔类型进行划分，还应结合化学特征和子实上层K反应进行综合研究。下一步应继续增加地图衣属物种序列，综合表型与基因型特征，进一步探讨该属的属下、种间的划分依据，以期建立更合理自然的分类系统。

3 检索表

世界 Badioatrum group 物种检索表

1.地衣体棕色，裂片表面凹陷呈脐叶状，有胶质层分布，髓层I–，孢子20–24 × 10–15 µm……………………………………………………………**R. rittokense** (Hellb.) Th. Fr.

1.地衣体裂片非脐叶状，表面平整或凸起呈球形……………………………………………2

2.地衣体表面有衣瘿分布（由第二光合共生物与共生真菌构成）............***R. hensseniae*** Brodo

2.地衣体表面无衣瘿分布………………………………………………………………………3

3.子囊孢子通常较小，长度为12–22 µm………………………………………………………4

3.子囊孢子较大，长度为22–42 µm……………………………………………………………8

4.髓层呈黄色，含地图衣酸..............................***R. flavomedullosum*** Elix & P.M. McCarthy

4.髓层白色，不含地图衣酸……………………………………………………………………5

5.髓层I–，子实上层深蓝色，K–，孢子较小，12–15 × 7–7.5 µm....***R. alaxense*** J.W. Thomson

5.髓层I+蓝色……………………………………………………………………………………6

6.地衣体含有布尔吉尼克酸，孢子长度为15–24 μm....***R. vigilans*** P.M. McCarthy & Elix

6.地衣体有或无三苔色酸，孢子较小，长度12–16 μm.......................................7

7.地衣体缺失或发育不良，子实上层棕黑色，子实层浅红色，孢子棕色，12.5–20 × 5–7.5 μm，不含化学物质..............................***R. umense*** (H. Magn.) A. Nordin

7.地衣体发育较好，子实上层棕色，子实层无色，孢子为橄榄绿或棕色，12.5–16 × 6.5–7.5 um，有或无三苔色酸....................................***R. simillimum*** (Anzi) Lettau

8.子实上层橄榄棕色至墨绿色，K–，分布可溶于K试剂的晶体.....................9

8.子实上层红棕色至深棕色，K+紫红色，无晶体分布….............................10

9.地衣体浅灰色至深灰色或深棕色，裂片直径约1 mm，表面无粉霜，有或无光泽，平整或呈球状凸起，髓层K+黄色，K±红色（含有斑点酸，±降斑点酸）...***R. copelandii*** (Körb.) Th. Fr.

9.地衣体深棕色，裂片直径可达1.5 mm，表面通常有灰色粉霜，无光泽，平整或略有凸起，连续分布，髓层 K+黄色，只含斑点酸...***R. jemtlandicum*** Malme

10.髓层 K+黄色，只含斑点酸，孢子 28–38 × 12–l8 μm...***R. cinereonigrum*** Vain.

10.髓层 K–，不含斑点酸..11

11.地衣体含有三苔色酸，孢子橄榄棕色，35–40 × 10–15 μm...***R. sinense*** Zahlbr

11.地衣体不含有三苔色酸..12

12.地衣体裂片连续分布，前地衣体通常分布在地衣体边缘，孢子较小，23–36 × 13–16 μm，含有地弗地衣酸或无化学物质...***R. badioatrum*** (Flörke ex Spreng.) Th. Fr.

12.地衣体裂片呈分散分布，前地衣体通常分布在裂片之间，孢子较大，(27–)32–42.5 × 12.5–17.5(–20) μm，含有巴巴酸..***R. sichuanense*** Y.M. Zhang, L. Hu & W.C. Wang

Key to species of Badioatrum group in the world

1.Thallus brown, composed of umbilicate areolaes with epinecral layer, medulla I–, ascospore 20–24 × 10–15 μm, containing barbatic acid..***R. rittokense*** (Hellb.) Th. Fr.

1.Thallus composed of plane or convex areolaes, never umbilicate....................2

2.Thallus cephalodiate (which composed of second photosynthetic symbiont and symbiotic fungus)...***R. hensseniae*** Brodo

2.Thallus not cephalodiate..3

3.Ascospores small, 12–22 μm long...4

3.Ascospores large, 22–42 μm long...8

4.Medulla yellow above, containing rhizocarpic acid...***R. flavomedullosum*** Elix & P.M. McCarthy

4.Medulla white, not containing rhizocarpic acid...5

5.Medulla I–, epihymenium black-blue, K–, ascospores 12–15×7–7.5 μm..............

…………………………………………………………………***R. alaxense*** J.W. Thomson

5.Medulla I+blue……………………………………………………………………6

6.Containing bourgeanic acid, ascospores 15–24 μm long…………………………… ………………………………………………………***R. vigilans*** P.M. McCarthy & Elix

6.Containing ±gyrophoric acid, ascospores small, 12–16 μm long……………………7

7.Thallus reduced, epihymenium black brown, hymenium pale red, ascospores brown, 12.5–20 × 5–7.5 μm, containing no substance…………………………………………………***R. umense*** (H. Magn.) A. Nordin

7.Thallus well developed, epihymenium brown, hymenium hyaline, ascospores olive-green or brown, 12.5–16 × 6.5–7.5 um, containing ±gyrophoric acid…………………………………………***R. simillimum*** (Anzi) Lettau

8.Epihymenium olive-brown to green-black, K–, containing crystals dissolving in K……………………………………………………………………9

8.Epihymenium red-brown to dark brown, K+ red, not containing crystals……………………………………………………………………10

9.Thallus pale grey to dark grey or dark brown, areolaes up to 1 mm diam, without pruina, dull or shiny, plane to bullate; medulla K+ yellow, K± red, containing stictic acid and ±norstictic acid…………………***R. copelandii*** (Körb.) Th. Fr.

9.Thallus dark brown, areolae up to 1.5 mm diam, with faintly grey pruinose, dull, plane to weakly convex; medulla K+ yellow, containing stictic acid only……………………………………………***R. jemtlandicum*** Malme

10.Medulla K+ yellow, containing stictic acid only, ascospores 28–38 × 12–l8 µm……………………………………………***R. cinereonigrum*** Vain.

10.Medulla K–, not containing stictic acid……………………………………11

11.Thallus containing gyrophoric acid, spore olive-brown, 35–40 × 10–15 µm……………………………………………………***R. sinense*** Zahlbr

11.Thallus not containing gyrophoric acid……………………………………12

12.Areoles continuous, prothallus present along the margin, ascospores smaller 23–36 × 13–16 µm, containing diffractaic acid or no substances…………………***R. badioatrum*** (Flörke ex Spreng.) Th. Fr.

12.Areoles more or less scattered, prolthallus present between the areoles, ascospores (27–)32–42.5 × 12.5–17.5(–20) µm，containing barbatic acid……………………***R. sichuanense*** Y.M. Zhang, L. Hu & W.C. Wang

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